

Claims:

1. The use of an activator-independent MurD enzyme in a screening assay to identify inhibitors of the enzyme, which assay comprises contacting the enzyme with a test compound
5 in the presence of enzyme substrates and appropriate buffers and detecting any modulation of enzyme activity by the test compound.
2. The use as claimed in claim 1 wherein the activator-independent MurD enzyme comprises an amino acid sequence wherein one or more of the amino acid residues G96,
10 A112, A116, V126, L129, M133, G296, P298 and V422 is a variant amino acid residue.
3. The use as claimed in claim 2 and wherein the activator-independent MurD enzyme comprises an amino acid sequence having one or more of the amino acid residues K96, C112, G116, T126, M129, L133, N296, S298 and I422.
15
4. The use as claimed in claim 1 and wherein the activator-independent MurD enzyme is an *E. faecalis* MurD enzyme.
5. The use as claimed in claim 1 and wherein the activator-independent MurD enzyme
20 comprises the amino acid sequence
MKKITTYQNK KVLVLGLAKS GVSAAKLLHE LGALVTVNDA KQFDQNPDAQ
DLLTLGIRVV TGGHPIELLD EEFELIVKNP GIPYTNPLVA EALTRKIPII
TEVELAGQIA ECPIVGITGT NGKTTTTTMI GLLLNADRTA GEARLAGNIG
FPASTVAQEA TAKDDLVMEL SSFQLMGIET FHPQIAVTN IFEAHLDYHG
25 SRKEYVAAKW AIQKNMTVED TLILNWNQVE LQTLAKTTAA NVLPFSTKEA
VEGAYLLDGK LYFNEEYIMP ADELGIPGSH NIENALAAIC VAKLKNVSNV
QIRQTLKNFS GVPHRTQFVG EVQQRIFYND SKATNILATE MALSGFDNQK
LLLLAGGLDR GNSFDELVPA LLGLKAIVLF GETKEKLAEA AKKANIELIL
FAENVQTAVT IAFDYSEKDD TILLSPACAS WDQYPNFEVR GEAFMQAVQQ
30 LKESEM or a sequence having at least 85% sequence identity therewith.

6. The use as claimed in any preceeding claim and wherein the activator-independent MurD enzyme has up to 16 amino acids removed from the N-terminus, or up to 12 amino acids removed from the C-terminus, of the amino acid sequence.

5 7. The use as claimed in any preceeding claim and wherein the enzyme substrates are UDP-MurNac-L-Ala, D-Glutamate and ATP.

8. The use as claimed in any preceeding claim and wherein the enzyme is pre-incubated with the test compound before contacting it with the substrates.

10

15

20

25

30